

**What is claimed:**

1. A method for increasing production of alcohol from a saccharide source by an  
alcohologenic cell comprising,

5       contacting a saccharide source with an alcohologenic cell, and  
       exposing said cell to at least one compound of formula I,



wherein;

$R_1$  is H, OH or  $COOR_2$ ;

15  $R_2$  is H or alkyl;

R<sub>3</sub> is H, NH<sub>2</sub>, alkyl or alkenyl;

R<sub>4</sub> is H, alkyl, alkenyl, or a side chain of a naturally occurring amino acid; and

salts thereof;

20 wherein said exposing results in the increased production of alcohol by the  
alcohologenic cell as compared to a control.

2. A method for increasing growth of a cell comprising,

contacting a cell with a saccharide source, and  
25 exposing said cell to at least one compound of formula I,



wherein;

$R_1$  is H, OH or COOR<sub>2</sub>;

R<sub>2</sub> is H or alkyl;

35  $R_3$  is H,  $NH_2$ , alkyl or alkenyl;

R<sub>4</sub> is H, alkyl, alkenyl, or a side chain of a naturally occurring amino acid; and

salts thereof;

wherein said exposing results in the increased growth of said cell as compared to a control.

- 5     3.     The method of claim 1 or 2, wherein said compound of formula I is selected from the group consisting of lower aliphatic aldehydes, lower aliphatic  $\alpha$ -keto carboxylic acids, lower aliphatic dicarboxylic acids, amino acids, and salts of any of said acids.
- 10    4.     The method of claim 1, wherein said alcohol is ethanol and said alcohologenic cell is an ethanologenic cell.
5.     The method of claim 2, wherein said cell is an ethanologenic cell.
- 15    6.     The method of claim 4 or claim 5, wherein said cell is selected from the family Enterobacteriaceae.
7.     The method of claim 6, wherein said cell is *Escherichia* or *Klebsiella*.
- 20    8.     The method of claim 7, wherein said cell is a recombinant cell.
9.     The method of claim 8, wherein said cell is selected from the group consisting of *E. coli* KO4 (ATCC 55123), *E. coli* KO11 (ATCC 55124), *E. coli* KO12 (ATCC 55125), *K. oxytoca* M5A1, *K. oxytoca* P2 (ATCC 55307), and LY01 (ATCC \_\_\_\_\_).
- 25    10.    The method of claim 4, wherein said compound of formula I is selected from the group consisting of acetaldehyde, pyruvate, succinate, isocitrate, glutamate,  $\alpha$ -ketoglutarate, casamino acids, and yeast extract.
- 30    11.    The method of claim 10, wherein said compound of formula I is acetaldehyde.
12.    The method of claim 10, wherein said compound of formula I is pyruvate.
13.    The method of claim 10, wherein said compound of formula I is glutamate.
- 35

14. The method of claim 5, wherein said compound of formula I is selected from the group consisting of acetaldehyde, pyruvate, succinate, citrate, isocitrate, glutamate,  $\alpha$ -ketoglutarate, malate, casamino acids, and yeast extract.

5 15. The method of claim 14, wherein said compound of formula I is acetaldehyde.

16. The method of claim 14, wherein said compound of formula I is pyruvate.

17. The method of claim 14, wherein said compound of formula I is glutamate.

10

18. The method of claim 10 or 14, wherein said cell is exposed to glutamate and acetaldehyde.

15

19. The method of claim 10 or 14, wherein said cell is exposed to pyruvate and acetaldehyde.

20. The method of claim 10 or 14, wherein said cell is exposed to fumarate and malate.

20

21. The method of claim 10 or 14, wherein said cell is exposed to  $\alpha$ -ketoglutarate and succinate.

22. The method of claim 1 or 2, further comprising providing said cell in an aqueous solution.

25

23. The method of claim 1 or 2, wherein said saccharide source is selected from the group consisting of cellooligosaccharide, lignocellulose, hemicellulose, cellulose, pectin, xylose, glucose, and any combination thereof.

30

24. The method of claim 1 or 2, wherein said cell is exposed to said compound of formula I for a period of time between about 1 and about 96 hours.

25. The method of claim 1 or 2, wherein said method is performed at a pH between about 6 and about 8.

35

26. The method of claim 25, wherein said method is performed at a pH of about 6.5.

030624-01601

27. The method of claim 1 or 2, wherein said method is performed at a temperature between about 20° and about 40° C.

5 28. The method of claim 27, wherein said method is performed at a temperature of about 35° C.

29. The method of claim 1 or 2, wherein said compound is present at a concentration between about 0.1 and about 4.0 g/L.

10

30. The method of claim 1 or 2, further comprising exposing said cell to said compound more than once.

15

31. The method of claim 1 or 2, further comprising exposing said cell to two or more different compounds of formula I.

32. The method of claim 31, wherein said exposing of said cell to said compound is performed at time intervals between about 1 hour and about 24 hours.

20

33. The method of claim 1 or 2, further comprising agitating said cell, said saccharide source, and said compound between about 50 rpm and about 200 rpm.

34. The method of claim 2, wherein said increased growth is indicated by increased cell density or decreased cell replication time.

25

35. The method of claim 34, wherein said increased cell density is indicated by an optical density of between about 2 and about 3 at 550 nm after 24 hours.

30

36. The method of claim 4, wherein said increased production of ethanol is indicated by an increase in volumetric productivity.

37. The method of claim 36, wherein said volumetric productivity is between about 0.3 g/L and about 0.5 g/L

35

38. The method of claim 1 or 2, wherein said method is performed in a fermentor vessel.

39. The method of claim 38, wherein said cell and said saccharide source are provided in an aqueous solution.

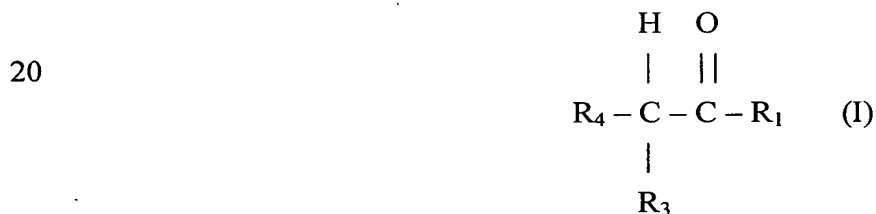
40. The method of claim 39, wherein said aqueous solution comprises a fermentation  
5 medium.

41. The method of claim ~~40~~, wherein said fermentation medium comprises Luria broth or CSL broth.

10 42. The method of claim ~~1~~ or 2, wherein said method is suitable for simultaneous saccharification and fermentation.

43. ✓ A growth medium suitable for use in an improved fermentation process comprising:

15 a saccharide source;  
a basal nutrient medium, and  
at least one compound of formula I,



wherein;

25  $\text{R}_1$  is H, OH or  $\text{COOR}_2$ ;  
 $\text{R}_2$  is H or alkyl;  
 $\text{R}_3$  is H,  $\text{NH}_2$ , alkyl or alkenyl;  
 $\text{R}_4$  is H, alkyl, alkenyl, or a side chain of a naturally occurring amino  
acid; and  
30 salts thereof.

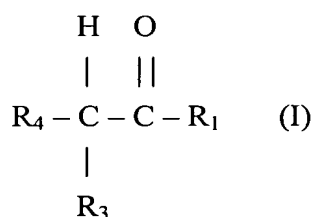
44. The growth medium of claim 43, wherein said saccharide source is selected from the group consisting of cellooligosaccharide, lignocellulose, hemicellulose, cellulose, pectin, xylose, glucose, corn steep liquor, and any combination thereof.

35 45. The growth medium of claim 43, wherein said basal nutrient medium is Luria broth or CSL broth.

46. The growth medium of claim 43, wherein said medium is suitable for use in simultaneous saccharification and fermentation.

47. The growth medium of claim 43, wherein said growth medium is packaged with instructions for use.

48. ✓ A fermentation reaction mixture suitable for producing ethanol comprising, a growth medium having a saccharide source, an ethanogenic cell, and an exogenous source of at least one compound of formula I,



wherein;

$\text{R}_1$  is H, OH or  $\text{COOR}_2$ ;

$\text{R}_2$  is H or alkyl;

$\text{R}_3$  is H,  $\text{NH}_2$ , alkyl or alkenyl;

$\text{R}_4$  is H, alkyl, alkenyl, or a side chain of a naturally occurring amino acid; and salts thereof.

49. The fermentation reaction mixture of claim 48, wherein said saccharide source is selected from the group consisting of cellooligosaccharide, lignocellulose, hemicellulose, cellulose, pectin, xylose, glucose, corn steep liquor, and any combination thereof.

50. The fermentation reaction mixture of claim 48, wherein said ethanogenic cell is from the family Enterobacteriaceae.

51. The fermentation reaction mixture of claim 48, wherein said reaction mixture is suitable for use in simultaneous saccharification and fermentation.

52. The growth medium of claim 43, wherein said compound of formula I is selected from the group consisting of acetaldehyde, pyruvate, succinate, citrate, isocitrate, glutamate,  $\alpha$ -ketoglutarate, malate, fumarate, a yeast extract, and a casamino acid.
- 5 53. The fermentation reaction mixture of claim 48, wherein said compound of formula I is selected from the group consisting of acetaldehyde, pyruvate, succinate, isocitrate, glutamate,  $\alpha$ -ketoglutarate, and a casamino acid.
54. The growth medium of claim 52, wherein said compound of formula I is selected  
10 from the group consisting of acetaldehyde, pyruvate, succinate, citrate, isocitrate, glutamate,  $\alpha$ -ketoglutarate, and malate.
55. The fermentation reaction mixture of claim 53, wherein said compound of formula I is selected from the group consisting of acetaldehyde, pyruvate, succinate,  
15 isocitrate, glutamate, and  $\alpha$ -ketoglutarate.

T051310 "4523230360